

COMPARATIVE ROOT C INPUTS UNDER A MIXED SWARD AND CONVENTIONAL RYEGRASS/CLOVER PASTURE

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Introduction

Soils contain the largest amount of carbon (C) in the terrestrial ecosystem with roughly twice the amount of C stored in the soil as found in the atmosphere (Batjes, 1996; Powlson et al., 2011). Consequently, relatively small changes to the soil C pool can influence the global C balance. Management practices that increase the soil C pool are being considered as an approach for sequestering C from the atmosphere.

Agricultural land covers approximately 40% of the global land surface (Smith et al., 2008) with grasslands and grazed land covering 70% of this land (Soussana and Luscher, 2007). The soil C pool of these agricultural ecosystems is primarily driven by the balance between C inputs and outputs (Paustian et al., 2000). Inputs to the soil C pool under these pasture systems are through plant biomass production (above and below-ground) and through returns of animal excreta while outputs are largely through microbial respiration. Increasing the inputs of C to a soil will generally have a positive effect on the soil C pool up until a C saturation point (Six et al., 2002). Soil C saturation is a soil's maximum ability to store C irrespective to changes in management (Six et al., 2002). While the surface of the soil may be near the C saturation point, the C content of soils usually declines with soil depth (Jobbagy and Jackson, 2000) and is likely to be further from C saturation. Thus, there is an opportunity to increase the C content of soils with depth by increasing the inputs of C further down in the soil profile.

There is significant interest in the role of roots to increase soil C under pastures. Roots provide a C input to the soil C pool through root mass, root turnover and rhizodeposition (Farrar et al., 2003). Studies have demonstrated that roots contribute more to soil C and subsequent stabilisation than above-ground biomass (Katterer et al., 2011; Kong and Six, 2010) with the mean residence time of root derived C in soil approximately 2.4 times that of aboveground C inputs (Rasse et al., 2005). Therefore, increasing the C inputs to soil through increasing root mass or the depth distribution of roots in the soil profile are potential strategies for increasing soil C.

Studies have shown that increasing species richness can increase the ecosystem productivity (Tilman et al., 1996) with the most diverse and productive plant communities displaying the greatest and deepest distributions of root biomass (Mueller et al., 2013). Therefore, it has been suggested that there is the potential to increase C sequestration through a combination of changes in biodiversity of a system or through land use change or management (Steinbeiss et al., 2008).

New Zealand agriculture is dominated by a ryegrass and clover based pasture, which is typically shallow rooting (Bolinder et al., 2002; Crush et al., 2005). Mixed sward pastures (including species such as chicory, lucerne and plantain) would be moderately diverse

compared to conventional ryegrass-clover swards. Studies of root mass that have been carried out in New Zealand (Dodd and Mackay, 2011; Matthew, 1996; Saggart et al., 1997) have focussed on ryegrass-clover systems. However, little is known about the rooting characteristics of these mixed swards in comparison to traditional ryegrass clover swards. Our objective was to determine whether a mixed sward pasture had greater root mass, rooting depths, and annual inputs of C to soil than a conventional ryegrass-clover pasture.

Methods

Site Description

The study site was located 7 km northeast of Hamilton in the Waikato region, North Island, New Zealand (37°46'13.62''S, 175°22'40.64''E) at Scott Farm. The soil type of the study location was classified as a Matangi silt loam (Typic Orthic Gley Soil) (Hewitt, 1993; Mudge et al., 2011).

The study site was an existing plant diversity trial operated by DairyNZ consisting of a series of 0.5 ha flat paddocks. These paddocks were rotationally grazed year round by herds with stocking rates and farm management similar to that used in commercially run dairy farms. The trial consisted of a series of treatments with differing pasture types. Each treatment had 3 replicates in a randomised block design. For the purpose of this study, only two pasture types were focussed on, a conventional ryegrass clover sward and a moderately diverse (mixed) sward. Ryegrass-clover refers to a ryegrass (*Lolium perrene*) and clover (*Trifolium repens*) pasture commonly used in New Zealand grazed systems. Moderately diverse ryegrass, or mixed sward refers to a ryegrass and clover pasture with the addition of the species, chicory (*Cichorium intybus*), plantain (*Plantago lanceolata*), and lucerne (*Medicago sativa*). These two pasture swards had an additional 3 paddocks each where the ryegrass component of each pasture was replaced with a high-carbohydrate ryegrass cultivar.

Sample collection

Root samples were collected four times through a year from both high and low sugar ryegrass cultivar treatments. Samples were taken in April 2012, July 2012, October 2012 and February 2013 (autumn, winter, spring and summer respectively). Ten sample sites in each paddock were located by generating random co-ordinates and ensuring no sample was taken within 5 m of any fenceline. At each core location, the aboveground species composition was determined using a 0.25 m² quadrant and recording the relative aboveground species abundance (%) within the quadrant. Soil cores (50 mm diameter, 300 mm depth) were taken from the centre of the quadrant and the core partitioned in to 3 depth increments (0-100, 100-200, and 200-300 mm) and kept separate for individual processing.

Root Biomass

Each depth increment from each core was washed with water, collecting all root material retained on a 250 µm sieve. Samples were first passed through a 2 mm sieve with water to loosen the soil particles ensuring all water was collected. Root material was floated off and collected using a 250 µm sieve. It is acknowledged that some very fine root material may have been lost through the 250 µm sieve. Root material was dried in a fan forced oven at 65°C for at least 48 hours before weighing to give root biomass.

Root turnover

Root turnover was not directly measured in this study, but estimates of turnover can be made as the difference in the maximum and minimum root masses within each pasture treatment during one year using a method modified from Scurlock et al., (2002). The difference of root

turnover between each treatment was then used to obtain the difference in root input between the species. The C input of this root turnover was estimated using a C content of roots of 40 % DM.

Root Parameter measurements

Root parameters were obtained following extraction of individual plants of each species in July 2012 and October 2013. Individual plants were isolated and extracted down to 30 cm keeping the bulk of soil intact around the root system where possible. Soil was gently shaken and washed from the root system using water, ensuring that the root profile remained as intact as possible. Roots were evenly spread in a water layer on a transparent tray (40 x 30 cm) and imaged at a resolution of 600 dpi (dots per inch) using an Epson Expression scanning system. Root images were analysed for total root length, average root diameter, projected and surface area, root volume and surface area using WinRHIZO software (Regent instruments). Root length and surface area was standardized using the dried root mass to give specific root length and specific surface area.

Calculations and Statistical Analysis

Root dry weights were converted to an equivalent mass per hectare of soil surface (kg ha^{-1}) using the cross sectional surface area of the soil core. Statistical analyses were performed using a two-way analysis of variance (ANOVA) for the root mass data using sward type and ryegrass cultivar as variables. The results of the two-way ANOVA showed that the sugar ryegrass cultivar had no significant effect ($P>0.05$) on root mass. The root mass data was pooled based on pasture sward type (mixed sward vs ryegrass-clover) and a one-way ANOVA performed. A repeated measures ANOVA on the seasonal root mass data was also carried out. Root mass data was log transformed to meet the assumptions of the ANOVA. Statements about significance of the results refer to $p<0.05$. ANOVA was performed using R v2.15.0 and repeated measures ANOVA performed using STATISTICA.

Root parameters (length and surface area) were standardised by root mass to give SRL and SSA and the root mass and volume was used to calculate root density.

Results

Root mass

Root mass displayed a seasonal pattern with the greatest mass between the summer/autumn period with lowest mass during winter. There was greater root mass under the mixed sward pasture compared to ryegrass clover for all seasons to 30 cm depth with average differences for the autumn, winter, and spring of 980, 1700, 700 kg ha^{-1} (Figure 1a). Summer had the greatest difference with 3700 kg ha^{-1} more root mass under the mixed sward ($p<0.05$). The 0-10 cm depth also had the greatest difference in the summer sampling with 2080 kg ha^{-1} more root mass in the mixed sward ($p<0.05$) (Figure 1b). There was significantly more root mass across all sampling dates for the mixed sward in the 10-20 cm depth ($p<0.05$) with about 1.5 to 2 times the root mass. Autumn and summer showed the greatest differences with 857 and 952 kg ha^{-1} greater root mass (Figure 1c). The 20-30 cm depth showed approximately double the root mass in the mixed sward compared to the ryegrass-clover. The greatest difference occurred in summer with 650 kg ha^{-1} root mass with winter and spring also displaying significantly more root mass ($p<0.05$) (Figure 1d).

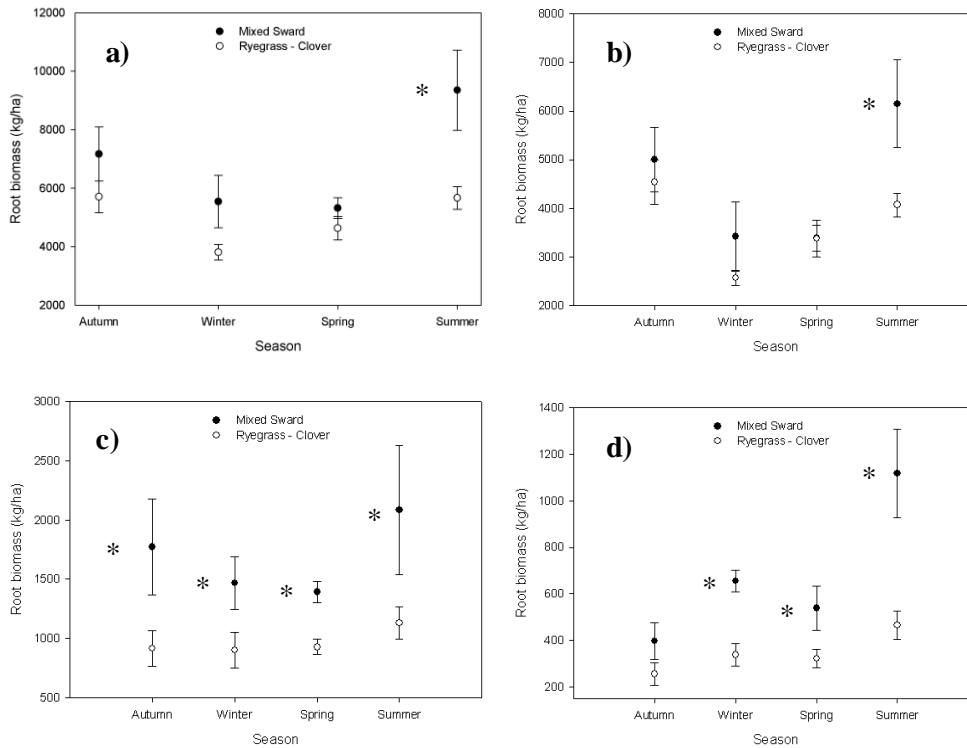


Figure 1 Root biomass for of the mixed sward (black filled circle) and ryegrass-clover sward (black outline, open filled circle) each season for the various soil depths, a) 0-30 cm, b) 0-10 cm, c) 10-20 cm, d) 20-30 cm. * represents a statistically significant difference between the two swards of $p < 0.05$.

Root turnover and C input

Estimated annual root turnover was higher in the mixed sward compared to ryegrass clover with a difference of 2134 kg ha^{-1} root mass to 30 cm depth (Table 1). Assuming the root material was approximately 40% C, this difference equated to a greater annual C input to soil of 853 kg C ha^{-1} under the mixed sward (0-30 cm). The C input was also greater in the 0-10, 10-20, 20-30 cm depths with 319, 184, and 204 kg C ha^{-1} under the mixed sward pasture respectively.

Table 1 Comparative root turnover (kg ha^{-1}) for the mixed sward and ryegrass-clover pastures, the corresponding difference and C input assuming a root DM C content of 40%.

	0-30 cm	0-10cm	10-20cm	20-30 cm
Mixed Sward	4027	2759	690	719
Ryegrass-Clover	1893	1963	229	210
Difference	2134	796	461	509
C input* (kg C ha^{-1})	853	319	184	204

* Assuming root DM was approximately 40% C

Root parameters

Specific root length (SRL) ranged from 1.91 cm g⁻¹ for lucerne to 67.80 cm g⁻¹ for ryegrass (Table 2). The larger tap rooted species (lucerne and chicory) had lower SRL whereas the finer rooted species (ryegrass, clover) had higher SRL. The species with larger SRL also showed greater specific surface area (SSA) and smaller average root diameter compared to the species with smaller SRL which showed lower SSA and greater average root diameter. Root density ranged from 0.10 g cm⁻³ for chicory to 0.22 g cm⁻³ for lucerne. Ryegrass, clover and plantain showed similar densities between 0.13 and 0.16 g cm⁻³.

Table 2 Root parameters for the individual plant species.

	Specific root length (cm g ⁻¹)	Specific Surface Area (cm ² g ⁻¹)	Root Density (g cm ⁻³)	Average Diameter (mm)
Ryegrass	67.80	674.44	0.13	0.32
Clover	47.54	510.04	0.15	0.34
Plantain	17.04	218.20	0.16	0.54
Chicory	2.92	62.69	0.10	0.91
Lucerne	1.91	44.67	0.22	1.10

Discussion

Root mass

The values obtained for root mass were consistent with other studies carried out on ryegrass-clover systems (Dodd and Mackay, 2011; Matthew, 1996; Saggar et al., 1997) and other forage species containing chicory and lucerne (Bolinder et al., 2002; Gentile et al., 2003). The mixed sward pasture had greater root mass compared to the conventional ryegrass-clover pasture for all samplings and depths.

The greatest differences in the root mass were generally observed during the summer sampling with lowest root mass in winter. This summer peak and winter low is also consistent with previous studies (Dodd and Mackay, 2011; Matthew, 1996; Saggar and Hedley, 2001) and was likely following peak pasture growth during spring followed by root/tiller death in autumn. It should be noted that the summer sampling coincided during a significant drought which may have contributed to the differences in root mass between the two pastures.

The mixed sward pastures consistently had greater root mass with soil depth (below 10 cm) throughout the year suggesting that the C input with soil depth may also be able to be increased. This increased rooting depth compared to the ryegrass-clover sward could be partly explained by an increase in species richness. Previous studies have demonstrated that diverse plant communities are associated with the deepest distributions of plant biomass (Mueller et al., 2013; Tilman et al., 1996). However, it is acknowledged that the ryegrass-clover and mixed sward pastures are very similar in terms species richness. Therefore, the increase in root mass cannot be solely attributed to the difference in species richness alone but may rather be due to the abundance of key species. Species such as lucerne and chicory have about 50% of roots in the top 30cm of the soil profile (Bolinder et al., 2002; Gentile et

al., 2003) compared to ryegrass where approximately 80% of root mass are found in the top 15 cm of soil (Bolinder et al., 2002; Crush et al., 2005). Thus, lucerne and chicory are likely to be the species contributing to the greater root mass with soil depth in the mixed sward pasture. Overall results were consistent with the mixed sward having greater root mass and greater rooting depth compared to the ryegrass-clover pasture.

Root turnover and C input

The root turnover was greater under the mixed sward pasture compared to the ryegrass-clover pasture. This greater root input leads to an opportunity to increase the C input to soil by approximately 853 kg C ha⁻¹ yr⁻¹. It is acknowledged that not all of the root turnover estimate would be stabilised in soil C but provides a relative comparison on differences between the root C input of the two pasture swards. The C input under the mixed sward pasture through root turnover was greater than the conventional ryegrass-clover pasture. This greater input at the 10-30 cm soil depth is likely to also be further from a C saturation limit compared to that seen in the 0-10 cm depth as was postulated by Dodd et al., (2011) as a potential strategy for increasing soil C in New Zealand Soils.

One limitation in the method used to estimate root turnover was that it fails to account for rhizodeposition. Rhizodeposition is the broad term used for a range of processes such as root exudates and cell sloughing where C enters the soil (Jones et al., 2009). The contribution of C released from roots through rhizodeposition has been estimated to be around 2.5 times the C incorporated in root mass (Johnen and Sauerbeck, 1977), though recent studies have shown about 50% of C allocated below ground remains in roots (Kuzyakov and Domanski, 2000; Kuzyakov and Schneckenberger, 2004).

Root parameters

The root parameters for the individual species found in these two swards demonstrated that the ryegrass and clover based pasture was composed of two species that typically have smaller root diameters and greater SRL than the mixed pasture and have a greater fine root mass. Root morphology may be more important in the stabilisation of C through their influence on C inputs and soil aggregation. Jastrow et al., (1998) demonstrated that roots contributed to the restoration of macroaggregate structure in a restored prairie where root density and root distribution can influence aggregate size and formation. Furthermore, root diameter is thought to influence the decomposition and turnover of roots where smaller fine roots (<2 mm) have a faster decomposition and turnover (Pacaldo et al., 2014). Therefore, the distribution of fine roots of the ryegrass-clover sward may be equally important as the increased root mass under the mixed sward in relation to the C input to soil. Thus, there is need for further investigation using more suitable techniques to better understand the C inputs under these two swards is needed.

Conclusion

This study has demonstrated that a mixed sward pasture had a greater root mass and greater rooting depth compared to a ryegrass and clover based pasture. This difference in root mass equated to a greater C input of around 853 kg/ha under the mixed sward pasture to 30 cm depth. However, individual root parameters suggest that the ryegrass and clover species have finer root systems than the other species found in the mixed sward which may also be of importance in terms of the overall C input to soil.

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